

## **Cell Synchronization Techniques:**

### **G<sub>1</sub>/S Synchronization:**

#### **For AD293 cells, and other p53 positive cells lines:**

Reagents:

L-mimosine stock solution (500mM)

(Stock solution is in 1xPBS + titrations of NaOH to facilitate L-mimosine solubility)

-L-mimosine is added to a final concentration of 250nM in media, mixed thoroughly and cells are incubated in T.C. incubator for 20-22hrs.

-To release cells, rinse 1x in 1xPBS, 1x in Growth media, and release in your normal volume of Growth Media.

#### **For HeLa cells – Double Thymidine Block:**

Reagents-

100mM Thymidine Solution (in dH<sub>2</sub>O)

Protocol-

- (1) Plate HeLa, and allow to grow to ~40% confluency
- (2) Add Thymidine to a final concentration of 2mM directly into media on plate, mix gently.
- (3) Incubate in T.C. incubator for 19hrs.
- (4) Aspirate media, rinse 2x with 1xPBS, 1x with Growth media.
- (5) Add Fresh Growth Media, return to T.C. incubator for 9hrs.
- (6) Add Thymidine at 2mM final concentration directly to media, mix gently.
- (7) Incubate in T.C. incubator for 16hrs.
- (8) Wash as above, and release cells into Fresh Growth Media for desired length of time.